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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/803,165	03/09/01	SOBEK	H 5328

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ROCHE MOLECULAR SYSTEMS INC
PATENT LAW DEPARTMENT
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EXAMINER	
NGUYEN,L	
ART UNIT	PAPER NUMBER
1635	6

DATE MAILED: 11/06/01

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary	Application No.	Applicant(s)	
	09/803,165	SOBEK ET AL.	
	Examiner	Art Unit	
	Lauren Nguyen	1635	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133)
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☐ Responsive to communication(s) filed on _____ .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-14* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-14* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s) _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

Priority

1. Acknowledgment is made of applicant's claim for foreign priority based on an application filed in EPO on 3/11/2000. It is noted, however, that applicant has not filed a certified copy of the EPO application as required by 35 U.S.C. 119(b).

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> *The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.*

2. Claims 1, 3, and 6-9 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claims 1, 3, and 6-9 recite a "...**thermostable** ... polymerase..." The term "thermostable" has not been defined in the specification or claims; the term "thermostable" is a relative word and without defining a reference temperature and corresponding enzymatic activity which define that term, one of skill in the art would not know the metes and bounds of the limitation recited in the term "thermostable."

3. Claims 3 and 5 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claims 3 and 5 recite the limitation of having "Y→..." mutation. There is insufficient antecedent basis for this limitation in the claim since there is no referral to the "Y" being mutated as being part of the Y-GG/A motif recited in claim 1.

4. Claim 12 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claim 12 recites the limitation of "...mutagenesis of **the** gene, followed by ... purification of **the** protein." There is insufficient antecedent basis for this limitation in the claim.

5. Claims 13 and 14 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Claims 13 and 14 provide for the use of "the polymerase", but, since the claim does not set forth any steps involved in the method/process, it is unclear what method/process applicant is intending to encompass. A claim is indefinite where it merely recites a use without any active, positive steps delimiting how this use is actually practiced.

Claims 13 and 14 are also rejected under 35 U.S.C. 101 because the claimed recitation of a use, without setting forth any steps involved in the process, results in an improper definition of a process, i.e., results in a claim which is not a proper process claim under 35 U.S.C. 101. See for example *Ex parte Dunki*, 153 USPQ 678 (Bd.App. 1967) and *Clinical Products, Ltd.* v. *Brenner*, 255 F. Supp. 131, 149 USPQ 475 (D.D.C. 1966).

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> *A person shall be entitled to a patent unless –*
>
> *(a) the invention was known or used by others in this country, or patented or described in a printed publication in this or a foreign country, before the invention thereof by the applicant for a patent.*
>
> *(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.*

6. Claims 1-5 and 9-13 are rejected under 35 U.S.C. 102(b) as being anticipated by Pisani *et al.* (Biochemistry, Vol. 37, p. 15005-15012, 1998).

Claim 1 is drawn to a thermostable mutant B-type DNA polymerase comprising a Y-GG/A amino acid motif between the N-terminal 3'-5' exonuclease domain and the C-terminal polymerase domain wherein the tyrosine motif is substituted with another amino acid. Claims 2-5 are drawn to the mutant polymerase of claim 1 wherein the tyrosine of the motif is substituted with an amino acid with an aromatic side chain or a hydrophilic side chain, or having a Y→F, Y→W, or Y→H mutation or having a Y→N or Y→S mutation. Claims 9-12 are drawn to the DNA, vector, transformed host cell, and process for obtaining the mutant polymerase of claim 1. Claim 13 is drawn to a method of using the mutant polymerase of claim 1 for synthesizing nucleic acids.

Pisani *et al.* discloses mutant B-type DNA polymerases isolated from the thermoacidophilic archeon, *Sulfolobus solfataricus*. As disclosed in the abstract, Pisani *et al.* discloses six site-specific mutations within the Y-GG/A motif located in the DNA pol region 1 (p. 15009, paragraph 2 and Figure 4A). Among the mutants disclosed in Pisani *et al.*, Tyr^{495}→Phe, Ser mutants were also disclosed (p.15009, paragraph 2 and Figure 4A). All of the polymerase mutants described in Pisani *et al.* were encoded in plasmid pT7polS, overexpressed in E. coli, purified, and subjected to DNA polymerase and 3'-5' processivity assays (p. 15007, paragraphs 1-5).

Therefore, the invention of the above claims is anticipated by Pisani *et al.*

7. Claims 1-5 and 9-13 are rejected under 35 U.S.C. 102(b) as being anticipated by Truniger *et al.* (Journal of Molecular Biology, Vol. 286, p. 57-69, February 1999).

Claim 1 is drawn to a thermostable mutant B-type DNA polymerase comprising a Y-GG/A amino acid motif between the N-terminal 3'-5' exonuclease domain and the C-terminal polymerase domain wherein the tyrosine motif is substituted with another amino acid. Claims 2-5 are drawn to the mutant polymerase of claim 1 wherein the tyrosine of the motif is substituted with an amino acid with an aromatic side chain or a hydrophilic side chain, or having a Y→F, Y→W, or Y→H mutation or having a Y→N or Y→S mutation. Claims 9-12 are drawn to the DNA, vector, transformed host cell, and process for obtaining the mutant polymerase of claim 1. Claim 13 is drawn to a method of using the mutant polymerase of claim 1 for synthesizing nucleic acids.

Truniger *et al.* discloses mutant B-type DNA polymerases derived from ϕ29 DNA polymerase, a eukaryotic B-type DNA polymerase. As disclosed on p. 59, paragraph 4 and in Figure 2, Truniger *et al.* discloses six site-specific mutants with substitutions in a Y-GG/A motif located between the ExoIII motif and the polymerase domain (see legend of Figure 2). Among the mutants disclosed in Truniger *et al.*, Tyr^{226}→Phe, Ser mutants were also disclosed (Figure 2). All of the polymerase mutants described in Truniger *et al.* were encoded in plasmids, overexpressed in *E. coli*, purified, and subjected to DNA polymerase assays (p. 67, paragraphs 1-7).

Therefore, the invention of the above claims is anticipated by Truniger *et al.*

8. Claims 1-5 and 9-13 are rejected under 35 U.S.C. 102(b) as being anticipated by Truniger *et al.* (EMBO, Vol. 15, p. 3430-3441, 1996).

Claim 1 is drawn to a thermostable mutant B-type DNA polymerase comprising a Y-GG/A amino acid motif between the N-terminal 3'-5' exonuclease domain and the C-terminal polymerase domain wherein the tyrosine motif is substituted with another amino acid. Claims 2-5 are drawn to the mutant polymerase of claim 1 wherein the tyrosine of the motif is substituted with an amino acid with an aromatic side chain or a hydrophilic side chain, or having a Y→F, Y→W, or Y→H mutation or having a Y→N or Y→S mutation. Claims 9-12 are drawn to the DNA, vector, transformed host cell, and process for obtaining the mutant polymerase of claim 1. Claim 13 is drawn to a method of using the mutant polymerase of claim 1 for synthesizing nucleic acids.

Truniger *et al.* discloses mutant B-type DNA polymerases derived from ϕ29 DNA polymerase, a eukaryotic B-type DNA polymerase. As disclosed in the abstract and in Figure 1, Truniger *et al.* discloses twelve site-specific mutants with substitutions in the Y-GG/A motif located between the ExoIII motif and the polymerase domain. Among the mutants disclosed in Truniger *et al.*, Tyr^{226}→Phe, Ser mutants were also disclosed (Figure 1). All of the polymerase mutants described in Truniger *et al.* were encoded in plasmids, overexpressed in *E. coli*, purified, and subjected to DNA polymerase assays (p. 3433, paragraph 1 and Table 1).

Therefore, the invention of the above claims is anticipated by Truniger *et al.*

9. Claims 1-14 are rejected under 35 U.S.C. 102(a) as being anticipated by Bohlke *et al.* (Nucleic Acids Research, Vol. 28, p. 3910-3917, 10/15/2000).

Claim 1 is drawn to a thermostable mutant B-type DNA polymerase comprising a Y-GG/A amino acid motif between the N-terminal 3'-5' exonuclease domain and the C-terminal polymerase domain wherein the tyrosine motif is substituted with another amino acid. Claims 2-5 are drawn to the mutant polymerase of claim 1 wherein the tyrosine of the motif is substituted with an amino acid with an aromatic side chain or a hydrophilic side chain, or having a Y→F, Y→W, or Y→H mutation or having a Y→N or Y→S mutation. Claim 6 is drawn to the mutant polymerase of claim 1 wherein its wild-type form is obtainable from *Euryarchae*. Claims 7 and 8 are drawn to the mutant polymerase of claim 1 wherein its wild type form is obtainable from *Thermococcus aggregans* or wherein the amino acid sequence of its wild type form is ≥80% homologous to the amino acid sequence of wild-type *Tag* DNA polymerase. Claims 9-12 are drawn to the DNA, vector, transformed host cell, and process for obtaining the mutant polymerase of claim 1. Claims 13 and 14 are drawn to a method of using the mutant polymerase of claim 1 for synthesizing nucleic acids or for PCR.

Bohlke *et al.* discloses mutant B-type DNA polymerases derived from the euryarachaeon *Thermococcus aggregans* polymerase. As disclosed in the abstract, Bohlke *et al.* discloses five site-specific mutants with substitutions in a Y-GG/A motif located between amino acid positions 387-390 (p. 3913, paragraph 1). Among the mutants disclosed in Bohlke *et al.*, Tyr^{387}→Phe, Trp, His, Asn, and Ser mutants were also disclosed (Figure 2). All of the polymerase mutants described in Bohlke *et al.* were encoded in plasmids pTYPol, overexpressed in *E. coli*, purified, and subjected to DNA exonuclease and PCR assays (p. 3911, paragraphs 4-9 and p. 3920, paragraphs 1-2).

Therefore, the invention of the above claims is anticipated by Bohlke *et al.*

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> *(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.*

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

10. Claims 1-14 are rejected under 35 U.S.C. 103(a) as being unpatentable over Bohlke *et al.* (Nucleic Acids Research, Vol. 28, p. 3910-3917, 10/15/2000), Truniger *et al.* (EMBO, Vol. 15, p. 3430-3441, 1996), and Pisani *et al.* (Biochemistry, Vol. 37, p. 15005-15012, 1998) in view of Niehaus *et al.* (Gene, Vol. 204, p. 153-159, 1997) and Bordo *et al.* (Journal of Molecular Biology, Vol.271, p. 721-729, 1991).

Bohlke *et al.* teaches mutant B-type DNA polymerases derived from the euryarachaeon *Thermococcus aggregans* polymerase. As disclosed in the abstract, Bohlke *et al.* teaches five site-specific mutants, including Tyr^{387}→Phe, Trp, His, Asn, and Ser mutants, in a Y-GG/A motif

located between amino acid positions 387-390 (p. 3913, paragraph 1). All of the polymerase mutants described in Bohlke *et al.* were encoded in plasmids pTYPol, overexpressed in *E. coli*, purified, and subjected to DNA exonuclease and PCR assays (p. 3911, paragraphs 4-9 and p. 3920, paragraphs 1-2).

Truniger *et al.* teaches mutant B-type DNA polymerases derived from ϕ29 DNA polymerase, a eukaryotic B-type DNA polymerase. As disclosed in the abstract and in Figure 1, Truniger *et al.* teaches site-specific mutants with substitutions, Tyr^{226}→Phe, Ser, in the Y-GG/A motif located between the ExoIII motif and the polymerase domain. All of the polymerase mutants described in Truniger *et al.* were encoded in plasmids, overexpressed in *E. coli*, purified, and subjected to DNA polymerase assays (p. 3433, paragraph 1 and Table 1). Furthermore, the study by Truniger *et al.* demonstrates that mutations in the Y-GG/A motif lead to phenotypes which possess altered polymerase/exonuclease balance. In particular, the Y226F mutant exhibits favorably high polymerization/exonuclease ratios. Moreover, using multiple sequence alignment, Truniger *et al.* shows that the "YxGG/A motif can also be found in nearly all eukaryotic-type DNA polymerases (p. 3431, paragraph 3)." After aligning 51 sequences from viral, bacterial, and TP-primed DNA polymerase and analyzing the infrequency of substitutions within the YxGG/A motif, Truniger *et al.* were able to define the YxGG/A motif as being "conserved for the whole family of eukaryotic-type DNA polymerases (p. 3431, paragraph 3)."

Pisani *et al.* teaches mutant B-type DNA polymerases isolated from the thermoacidophilic archeon, *Sulfolobus solfataricus*. As disclosed in the abstract, Pisani *et al.* teaches six site-specific mutations, including Tyr^{495}→Phe, Ser mutants, within the Y-GG/A motif located in the DNA pol region 1 (p. 15009, paragraph 2 and Figure 4A). All of the

polymerase mutants described in Pisani *et al.* were encoded in plasmid pT7polS, overexpressed in *E. coli*, purified, and subjected to DNA polymerase and 3'-5' processivity assays (p. 15007, paragraphs 1-5).

Niehaus *et al.* teaches the sequence and cloning of a recombinant (removal of 3 intein sequences) thermostable polymerase isolated from *Thermococcus*. As disclosed in the abstract, Niehaus *et al.* teaches that the resultant polymerase shows DNA polymerase and 3'-5' exonuclease activity at high temperatures and is a "promising candidate for use in PCR."

Bordo *et al.* teaches amino acid substitutions in site-directed mutagenesis which would "least likely to disturb protein structure, either locally or in its overall folding pathway, and most likely to allow probing of the structural and functional significance of the substituted site (see abstract)." As listed in the matrix depicted in Figure 1C, favorable substitutions for tyrosine include histidine (+8), phenylalanine (+23), tryptophan (+12).

One of ordinary skill in the art would have been motivated to make a thermostable mutant DNA polymerase comprising a tyrosine substitution in the Y-GG/A motif since said substitution was taught by Bohlke *et al.*, Truniger *et al.* and Pisani *et al.* Furthermore, by combining the teachings of Truniger *et al.* and Niehaus *et al.*, one of ordinary skill in the art would have been motivated to make and use the claimed thermostable mutant DNA polymerases in PCR methods since Truniger *et al.* teaches that the YxGG/A motif is conserved for the whole family of eukaryotic-type DNA polymerases and, in ϕ29 DNA polymerase, the Y226F mutant exhibits high polymerization/exonuclease ratios while Niehaus *et al.* teaches that recombinant *Thermococcus* DNA polymerase is a promising candidate for use in PCR.

It would have been obvious to one of ordinary skill in the art to make a thermostable mutant DNA polymerase comprising a tyrosine substitution in the Y-GG/A motif since said substitution was taught by Bohlke *et al.*, Truniger *et al.* and Pisani *et al.* Moreover, using the combined teachings of Truniger *et al.* and Niehaus *et al.,* it would have been obvious to one of ordinary skill in the art to make and use the claimed thermostable mutant DNA polymerases in PCR methods since Truniger *et al.* teaches that the YxGG/A motif is conserved for the whole family of eukaryotic-type DNA polymerases and, in ϕ29 DNA polymerase, the Y226F mutant exhibits high polymerization/exonuclease ratios while Niehaus *et al.* teaches that recombinant *Thermococcus* DNA polymerase, a eukaryarchae-type DNA polymerase, is a promising candidate for use in PCR. Furthermore, since Bordo *et al.* teaches histidine, phenylalanine, and tryptophan as favorable substitutions for tyrosine, it would have been obvious to one of ordinary skill in the art to make DNA polymerase mutant containing such substitution for tyrosine in the conserved Y-GG/A motif.

One of ordinary skill in the art would have had a reasonable expectation of success in making a thermostable mutant DNA polymerase comprising a tyrosine substitution in the Y-GG/A motif since said substitution was taught by Bohlke *et al.*, Truniger *et al.* and Pisani *et al.* Moreover, by combining the teachings of Truniger *et al.* and Niehaus *et al.,* one of ordinary skill would have had a reasonable expectation of success in making and using the claimed thermostable mutant DNA polymerases in PCR methods since Truniger *et al.* teaches that the YxGG/A motif is conserved for the whole family of eukaryotic-type DNA polymerases and, in ϕ29 DNA polymerase, the Y226F mutant exhibits high polymerization/exonuclease ratios while

Niehaus *et al.* teaches that recombinant *Thermococcus* DNA polymerase, a eukaryarchae-type DNA polymerase, is a promising candidate for use in PCR.

Therefore, the invention of the above claims would have been *prima facie* obvious to one of ordinary skill in the art at the time of filing.

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Lauren Nguyen, Ph.D. whose telephone number is 703-308-0256. The examiner can normally be reached on Monday-Friday 9-5.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, John LeGuyader can be reached on 703-308-0447. The fax phone numbers for the organization where this application or proceeding is assigned are 703-308-4242 for regular communications and 703-305-7939 for After Final communications.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-308-0196.

Lauren Nguyen, Ph.D.
October 31, 2001

SEAN McGARRY
PRIMARY EXAMINER